

***Montana Academy of Sciences  
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**Abstracts**

# Oral Presentations

## Stress Affects on Swine

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**Introduction:** Stress status affects a sow's ability to be a productive mother. The three branches of productivity are: conception rate, Number Born Alive (NBA) and Average Daily Gain (ADG). Year one focused on calculating productivity and matching it with a stress status to determine a correlation. Year two compiles more data, but with an added concept- conception rate. Conception rate is often over looked because it is not quantifiable. Conception rate is the most important because productivity can not be measured if there are no offspring. This study shows that stress carrier swine cannot conceive as easily and are therefore less fertile. 2 out of the 5 stress carriers initially conceived. This is just the start of stress carriers troubles because they lack litter size, milk production and overall SPI. Also, 100% sows that did not conceive post farrowing were stress carriers.

### Methods:

- 1) Test artificially inseminated sows for PSS via ear prick, blood card.
- 3) As pigs farrow, conduct tests for pre farrowing conception rate, parity, litter size, initial litter weight, weaning weight and post farrowing conception rate. Come back into her heat cycle five days after weaning. By examining the sows after weaning it can be determined if their heat cycle is efficient.
- 4) Calculate SPI.

### Results

#### Sow Productivity Index

Genotype	2010	2011
NN	115.78	114.64
Nn	87.32	62.39

$$SPI = 100 + \{ 6.5 ( L-l ) \} + \{ 1.0 ( W -w ) \}$$

**Conclusions:** Sows that are stress carriers are negatively affected by PSS. 100% of the sows that did not initially conceive for the year 2011 were stress carriers. Stress negative swine, on average, have more piglets per litter. Stress negative swine had 9.14 piglets per litter whereas stress carriers had 4.25 piglets per litter. A 5 pig difference. Sows that were unaffected by PSS had a greater milk production. Litters that were stress negative gained 3.29 lbs. per day, but heterozygous litters gained 1.39 lbs. per day. A difference of almost 2lbs. per day per litter. Lastly, sows without PSS had higher SPIs. Sows without the stress gene had an average SPI of 110.81 while pigs that were PSS affected had an average SPI of 62.39. The industry average SPI is 100. Stress negatives were 10.81 above the industry standard while stress carrier sows were 37.61 units below the industry standard. With a difference of 48.42.

# **Comparision of Intestinal Parasite Percentages in Canines**

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**Introduction** Intestinal parasites are a common nuisance in canines. Roundworms are one of the most commonly seen parasites. However there are other parasites such as Giardia and Coccidia that can be common. Symptoms of intestinal parasites include; pot-bellied, poor growth, dull coat, diarrhea and vomiting, expelled worms in stool and vomit, weight loss/difficult weight gain, watery stool. A parasite can be transmitted; through the mother's milk if the mother is contaminated, contact of contaminated soil and feces, prenatal infection, other older dogs and puppies which carry parasites. The environment and health care provided contributes greatly to an animals parasite load. "Puppy mills" have been long time suspects for unhealthy environments and treatment. Therefore there would be a large suspicion that "puppy mills" have a higher parasite load then those dogs found elsewhere. The purpose of this project is to compare parasite percentages of different sites to find a correlation between them, the percentages and "puppy mills."

**Methods** Samples of dog feces will be gathered from three locations in Missoula, Montana include the Humane Society of Western Montana, Quick Paws and a pet store. At each site 20 random samples were collected in a plastic bag and labeled with the sample number. The date and site location were also noted. Testing for intestinal parasites can be done by performing fecal flotation. The test is performed by taking a small amount of the sample and putting it in a small cup then adding fecal solution to the feces and mixing them together. Next a test tube is filled with the solution and placed in a centrifugation for five minutes. Lastly the test tube is filled with fecal solution to the top and sits for eight minutes and then examined under a microscope.

## **Results**

### **Total Parasites Found Percentage**

No Parasites    Parasites

68                    32

**Conclusion**The results show a varied percentage and parasite type per location. Both the Humane Society and Quick Paws were relatively low with 10% and 15% parasites discovered in samples. The pet store which purchases their animals from "puppy mills," had a much greater parasite load at 70%. Therefore the suspicion is correct in this study that "puppy mills" generally produce less healthy animals. These are variables that must be considered the Humane Society treats their dogs for worms regularly to prevent the spread of intestinal parasite. Quick Paws also contains a low percent of parasite. People who use boarding kennels probably provide excellent care for their animals. Animals at the pet stores are usually puppies and younger animals generally have larger parasite load.

# **Difference of Particulate Matter at Roundabouts and Stop-Controlled Intersections**

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**Introduction:** The Missoula, MT valley is known to fail certain clean air standards. This poor air quality relates to inversions during the winter months, which are a result of the mountain-valley topography surrounding the city. Health officials know exposure to suspended particulate matter can irritate or sometimes lead to many respiratory problems and heart disease. Particles with a diameter less than 2.5 micrometers ( $PM_{2.5}$ ) are the focus of this study due to more dangerous effects caused by the ability to get deeper into the lungs. The City of Missoula has attempted to reduce pollutants such as  $PM_{2.5}$  through the establishment of roundabouts on heavily used and unsafe intersections. This project is designed to determine if in fact air pollution is reduced by the installation of roundabout intersections. To gather comparable data, two intersections were chosen nine blocks apart along the same major avenue, one stop-controlled intersection being Higgins-South Avenue, and the other intersection being the Higgins-Beckwith roundabout.

**Methods:** Data was collected by instruments locked in a car with attachment hoses outside the window for intervals of 4, 5, and 6 hours, and data logging intervals of 1 minute. DustTraks from TSI Inc. were used, gathering data on  $PM_{2.5}$  levels near the intersection in question.

**Results and Conclusions:** Comparing the two intersections, the Higgins-Beckwith roundabout had, on average, slightly elevated  $PM_{2.5}$  levels compared to the Higgins-South Avenue intersection. Values for the two intersections are as follows: the Higgins-Beckwith roundabout  $PM_{2.5}$  average being 61 micrograms per cubic meter ( $\mu\text{g}/\text{m}^3$ ), and the Higgins-South Avenue intersection  $PM_{2.5}$  average being  $55\mu\text{g}/\text{m}^3$ . The Missoula Urban Traffic Count Program 2010 study states the Higgins-Beckwith roundabout receives 10,300 average daily traffic while the Higgins-South Avenue intersection receives 9,500 average daily traffic, a difference of an average 800 vehicles per day. This difference could account for the  $PM_{2.5}$  level elevation at the roundabout. Despite these slight differences, both intersections registered “unhealthy for sensitive groups” on the EPA’s air quality index for the mean  $PM_{2.5}$  values during this study.

At the present state of this research, no conclusive evidence has been found which supports either intersection type having improved air quality. This is a result of a limited number of data gathering events, as well as a limited number of data gathering hours. Although generally higher  $PM_{2.5}$  levels were detected at the roundabout, further testing needs to be done to further distinguish between the two intersection types. To address these shortcomings, further research is designed to allow more extensive simultaneous monitoring of both sites.

# Identifying Species and Genus of Freshwater Sponges Inhabiting Blanchard Lake

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**Introduction** There are 3,000 marine sponges in the oceans. However, there are about 150 freshwater sponges; only 30 of the freshwater sponges are found belonging to the Spongillidae family, (The Xerces Society). The sponges' colors can either be yellow, gray, green, or a brownish color. They are mostly found on submerged objects such as rocks, pebbles, aquatic plants, logs, twigs, or branches. The sponges' can take many different forms. These forms can be a flat carpet-like sponge or even take a form as finger-like or branch-like shape. Sponges have simple bodies with no organs or differentiated tissues however the freshwater sponges are composed of small needle-like microscopic spicules which support the sponges' tissues. When the sponges are ready to reproduce, the sponges go through a process called gemmulation. These so called "gemmules" are expelled from the adult sponge to begin forming another sponge of the same type. In order to identify the sponges' genus and species, one must look at the spicules, a long spike-like needle, and afterwards one will look at the gemmules to identify the species of the sponge type, (Washington State Department of Ecology | Home Page | ECY WA DOE).

Are there multiple or a single sponge species in Blanchard lake. This allowed me to look at all 16 slides, two slides for each sponge, and see what their spicules and gemmules look like. Next, I compared my drawings to those of Pennak's Freshwater Invertebrates chart to see what the names of the spicules and gemmules I found within the sponges. In my research right now, I am able to identify each sponge's genus based on the details of the spicules. However, as I am looking at the details of the spicules, I have noticed that each spicule is different from each other. Because of this difference in the spicules, I have to look at the spicules more closely and narrow down the genus. With this, I will be able to isolate the main genus for each sponge sample. With future research, I am going to build a replica sponge model and place it where the Clearwater River junction runs into Blanchard Lake. I want to see why some sponge colonies like to colonize in deep murky water where the water hardly moves, and why other sponges prefer to be in the main current of the Clearwater River junction? With this future questions in mind, it will allow me to have a more of a understanding of why these sponges colonize in various of locations in the lake and take on these different forms instead of all being just one form.

## Methods and Materials

### Rapid Slide Mount:

- 4 or 5 mm of the sponges tissue
- 7- 10 Glass Slides
- 2 droplets of 2 molars of Concentrated Nitric Acid
- Mounting Medium
- Glass cover slip
- Heat Source

I collected eight sponges at Blanchard Lake, Montana by the Clearwater River junction. Once the samples were collected, they were placed in a container and are kept in a cool place to make sure that the gemmulation stage does not occur quickly before the actual test. For one to begin the identification of the sponges' genus and species, one must look at the spicules and gemmules to see what genus and species each sponge belongs to. The first is the spicule identification. First, one must put on an apron, safety goggles, and gloves to protect the hands from the concentrated nitric acid. Next, one will take a razor blade and cut off about 4-5 mm of the sponge sample, and utilize tweezers to place the sample on a glass slide. Next, one would use a pipette and add about 2-3 droplets of concentrated nitric acid onto the sample. Once this process is completed, light the mounting medium or any kind of heat source that is available. Then put the glass side between clippers which then one will gently go back and forth over the flame. This process will evaporate all the nitric acid away only leaving a small sample of the spicules. Finally, when the whole process is completed use a 4X- 100 power microscope, put it at the 2nd highest or the 3rd highest magnification, to view the details of each spicule. The second is the gemmule identification. Doing the same procedure as above, one will be able to see what the gemmules look and will be able to identify what the species is for each sponge sample.

**Results** In my first table, I examined the spicules of each sponge sample to determine the genus of each sponge. As a result, sponge samples #1, 3, 7, and 8 are the genus *Spongilla*. Whereas sponge samples #2, 4 and 6, are genus *Eunapius*. In my second table, I examined the gemmules of each sponge sample to determine the species of each sponge. The first time I did the examination of gemmules, I came across varieties of different shapes that were arranged like an hourglass or an orange slice. With the second, examination, I was able to find more gemmules with less diatom shells compared to the first examination. As a result, sponge samples # 2, 3, 4, 5, 7, and 8 resulted in the species *Eunapius*, and sponge samples # 1 and 6 are the species are *Duosclera*. In my final table, I name all the samples together to form the species name which resulted in the following: The species name for sponge sample # 1 is *Spongilla duosclera*. Sponge samples # 2 and 4 are *Eunapius eunapius*. For sponge samples # 3, 7, and 8, their species name is *Spongilla eunapius*. Sponge sample # 6 is the species name *Eunapius duosclera*. Lastly, sponge sample #5 is unidentified.

**Conclusion** After examining both the gemmules and spicules of all eight sponges, I am able to identify both the species and genus of the sponges. After re-examining the spicules, I came across what appeared to be a gemmule. The gemmules didn't appear on the Pennak's Invertebrates chart. After consulting with Dr. Addis, professor of biology at Carroll College, he explained that what I observed were not gemmules, but rather diatoms shells. After doing a second examination of the gemmules of all eight samples, I was able to find more gemmules than the first examination. As a result, I found that all the samples had *Eunapius* as the species, however for samples # 1 and 6, I did not find *Eunapius* for the species, but rather I found *Duosclera* as the species for sponge samples # 1 and 6, and lastly sponge sample # 5 is still unidentified because the genus is still yet to be determined. For my final experiment, I examined the spicules. With this, I narrowed down the genus of each sponge sample to either *Spongilla lacustris* or *Eunapius fragilis*. I looked at each picture of each sample and put the magnification to the 3rd highest power to look at the details of the spicules. I saw that most of the spicules were *Spongilla lacustris*. *Spongilla lacustris* appeared to have sharp ends with spines, whereas the other samples observed were identified as *Eunapius fragilis*, having blunt, spiny gemmoscleres. For sponge sample five, it was difficult to tell what the genus is because when I looked under the microscope, both *Spongilla* and *Heteromeyenia* were so closely related, I couldn't decipher the main genus of the sponge. Once I had both the species and genus of all the sponge samples, I then went onto naming the species name for each sponge. The species name for sponge sample # 1 is *Spongilla duosclera*. Sponge samples # 2 and 4 are *Eunapius eunapius*. For sponge samples # 3, 7, and 8, their species name is *Spongilla eunapius*. Sponge sample # 6 is the species name *Eunapius duosclera*. Lastly, sponge sample #5 is unidentified because its genus cannot be determined at this time. species name is In future research, I'll use this data and map out where and why different sponge species prefer to live in different parts of Blanchard Lake. However, in order to do this next step, I'll need to go back out to Blanchard Lake and scuba dive to all the sponge sites in the spring. This way, I can construct a replica model of a sponge and put it where the Clearwater River Junction comes into the lake. Also, I want to know why they tend to spread out to different parts of the lake instead of concentrating in one area.

# **Species Composition and Abundance of Turtles at Restored versus Natural Wetlands in the St. Lawrence River Valley of New York State**

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*1. University of Great Falls, MT*

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## **Introduction**

*Over the last several decades, wetlands have declined precipitously in North America due to drainage and land conversion to agriculture and other land uses. Programs such as the USDA National Resource Conservation Service's Wetland Reserve Program (WRP) and the US Fish & Wildlife Service's Partners for Fish and Wildlife Program (PFWP) restore wetlands with the goal of reestablishing ecosystem services and habitat for wildlife. Little is known about whether WRP or PFWP restorations are similar to natural wetlands in terms of functioning and biodiversity. The objective of this study was to test this by sampling turtle diversity at 31 restorations and 16 natural wetlands in the St. Lawrence Valley of New York, to compare relative abundance and diversity of turtles.*

## **Methods**

*Hoop trapping was performed for 30 trap/nights at each site in the month of June.*

## **Results & Conclusion**

*We detected painted turtle, snapping turtle, and Blanding's turtles at both restorations and natural locations. We found that wetland type is a significant predictor for both trap success and the abundance of snapping turtles. The proportion of male snapping turtles was higher in natural wetlands, while the proportion of female snapping turtles was higher in restorations. Turtles were similar in age at both wetland types. Therefore, we conclude that turtle community structure is similar in restored and natural wetlands, and that turtles may be a good indicator for well-functioning restorations.*

# ***A Novel Ligand-Receptor Interaction in the Activin Signaling Network in Drosophila***

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*Cells must communicate to ensure proper growth and development. This communication, or signaling, occurs when ligands bind receptors. Related ligands can bind related receptors, called isoforms, that can be encoded by a single receptor gene. One signaling mechanism, in which multiple ligands signal using different isoforms can be seen in the Activin pathway of Drosophila. Previous work has shown that Activin ligands have different binding and signaling affinities for the different isoforms of the Activin receptor, but not all ligand-receptor isoform signaling pairs have been identified. By conducting wing assay experiments that over-express different combinations of ligands and receptors, we show that one ligand, Activin- $\beta$ , can signal via one form of the Activin receptor Baboon (Baboon B), but not via the other two isoforms (Baboon A or C). Baboon B is expressed in the brain and the wing, which are also the two tissues where Activin- $\beta$  is known to signal.*

# **Generalizing Affine Constrained Optimization**

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## **Introduction:**

*The traditional study of the theory and applications of the canonical primal maximization and the dual minimization affine constrained optimization problems have been well studied over the last hundred years. This traditional study has been very basis and coordinate dependent, as is natural for the applications to real world settings. In this proposed research we want to abstract and generalize these situations to abstract vector spaces of arbitrary dimension. We would like to see how much generalizes to linear and affine (possibly continuous) transformations for arbitrary dimensions (e.g. Banach Spaces). We would like to see which features of classical affine programming can be generalized to this potentially infinite dimensional setting, and what hypothesis if any are necessary for these generalizations to hold.*

**Methods:** *We investigate the classical study of affine finite dimensional linear programming using Tucker tableaux and identify the features which are most fundamental to these problems. We use the language of functional analysis in order to describe a primal and dual affine program, and describe the analogues of known features of classical affine programming.*

**Results and Conclusion:** *We see that the dual program of a primal maximization affine program takes place in the vector space duals of the original primal affine transformation of vector spaces. We show how one can meaningfully describe feasible solutions of a primal-dual affine program relative to specified (closed) convex cones in each vector space. We then show that the **Tucker duality equation**, one of the cornerstones of affine programming, generalizes to this setting.*

# ***A Comparison of Growth and Yield of Fidel Butterhead Lettuce using a Hydroponic System, an Organic Soil, and an Inorganic Soil***

*Rachel Dickson*

*Big Sky High School, Missoula MT*

***Introduction*** *This project examines what type of growing system produces the healthiest Fidel Butterhead lettuce plants with the largest yield of leaves. In this project the different growth systems tested are a recirculating reservoir hydroponic system, an inorganic fertilized soil, an organic soil, and a control consisting of potting soil without any additives. The project examines which system works the best, and which fertilization technique is the most efficient for indoor plant growth.*

***Methods*** *The lettuce was planted in three different soil beds and in one recirculating hydroponic system. The plants were all measured twice a week, with weekly PH measurements. A two part nutrient solution was added to the hydroponic system in different ratios according to weekly stage of growth.*

***Results*** *The hydroponic system grew the fastest, with the highest number of leaves throughout, and the tallest plants. The inorganic grew the second fastest and had the second healthiest plants. The organic garden did worse than the control, neither ending with a significant plant height or a large yield of leaves.*

***Conclusions*** *The hydroponic growing system was the most efficient method. The inorganic soil was almost as effective as the hydroponic system, but did not produce as healthy plants. The inorganic garden did the worst, and the control did a little bit better than the inorganic garden. Overall, out of the gardening systems tested, the hydroponic system worked the best. It was not necessarily cost effective for small scale indoor growing, but definitely produced the healthiest, fastest growing lettuce plants.*

## ***Computational studies on anti-tumor AIMs***

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**Introduction:** Substituted anthracenyl isoxazole amides (AIMs) have a variety of medicinal properties including fluorescence and anti-tumor properties. The fluorescence, in part, could be due to their planar aromatic moieties and from their lipophilicity. The anti-tumor properties are in large part due to the planarity of the anthracene ring and its ability to pre-organize G-quadruplex (G4) conformation of DNA. The substitutions on the C-2 and C-10 carbons of the anthracene with either electron donating or electron withdrawing groups gives different binding affinity based on docking studies. A novel series of anthracenyl isoxazole amides (AIMs) have been synthesized, as well as, docking studies and will be described. Evidence consistent with a mechanism of action via the interaction of these compounds with G-quadruplex (G4) DNA and a structural based rationale for the observed selectivity of the AIMs for G4 is presented.

**Methods:** From previous studies on structure activity relationship (SAR) of AIMs, it became apparent that in each example examined, the presence of two dimethylaminopropyl groups, or “double tails,” led to increased efficacy. The hypothesis was that enhanced activity was attributed to increased bioavailability arising from the lowered lipophilicity, and another point to note out is that C(10) groups bearing lone pairs, i.e. chloride, appeared to be superior as well. Although, these two factors combined had not been examined, until the present work.

**Results:** The use of SNB-19 as a pre-screen is a good indicator of anti-tumor activity in the NCI 60, and represents the most active anti-tumor compound in this series to date. The overall mid-graph mean point (which approximates the average - log GI<sub>50</sub>) of the “double tail” compound is 5.71, which compares favorably to several agents currently used in general medical practice, such as fluorouracil (3.5), bleomycin (5.2) and rubidazole (5.6). A new working hypothesis has been developed using the most active of the AIMs docked with the most recent G4 crystal structure, pdb accession 1NZM, and will be described.

**Conclusions:** This study illustrates the use of SAR to guide the design of pharmacokinetic properties relatively early in the discovery process. Clearly, the next hurdle of this class of anticancer agents must pass is on the basis of its structural similarity to some polynuclear aromatic hydrocarbons, and thus potentially metabolic toxicity, to which our counter gambit is the proposition that the isoxazole moiety presents a metabolic CYP450 site, that is built to metabolize on demand. We will address this issue in forthcoming synthetic methodology and metabolism manuscripts and will report on our progress in due course.

# **The Effects of Fatigue on Muscle Force Generation in Females**

Michaela Fiore

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## **Introduction**

*Exercise-related fatigue is a normal challenge for every athlete. The amount of fatigue may vary due to the muscles targeted during the exercise, the physical capacity of the individual, and the duration of the exercise. The purpose of my project is to determine the average percentage of muscle force generation lost due to fatigue in the four major muscles in the leg (Quadriceps, hamstrings, gluteus medius, gastrocnemius) in females.*

## **Methods**

*To test the rate of fatigue, the subjects will complete two full circuits involving cardio vascular exercises as well as muscle strengthening exercises. By measuring the subjects' force generation with a handheld dynamometer in their four main leg muscles before testing, during testing, and after testing, I will determine the amount of muscle force generation lost due to fatigue. After testing, the subjects' data will be anonymously recorded and compared to other subjects' results as well as averaged into the total study's results.*

## **Results**

*Based on the results of the tests for all ten subjects, muscle force generation decreases in a very linear fashion corresponding with the amount of activity the subject is put through. The muscle that lost the most force generation was the quadriceps with 22% muscle force generation lost from the first test to the last test. The Hamstrings and Gastrocnemius lost the same percentage of muscle force generation from the first test to the last test with 20%. The Gluteus medius lost the least force generation from the first test to the last test with a total of 18% force generation loss.*

## **Conclusion**

*Muscle force generation decreased in a linear fashion throughout the testing. The only muscle not following the linear trend was the gluteus medius that followed a "V" shaped trend. The force generation decreased from the first to second test and then from the second test the force generation increased. With further reading and more testing a conclusion will be made about this interesting trend. The other muscles followed the hypothesized linear trend decreasing with fatigue.*

# **Effects of Octopamine on The Aggression and Courtship Behaviors of Male *Drosophila*.**

Austin Herron

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**Introduction:** To find out if a male *Drosophila* with low levels of octopamine are able to compete with male control flies who have normal levels of octopamine in a social courtship environment. Then in post copulation, noticing any habits of aggressive behavior exhibited by either the octopamine low, or octopamine normal fly.

**Methods:** Isolate both control, octopamine low, and female flies in separate isolation vials, then after 5-7 days, prepare the behavior room to the *Drosophila*'s comfortable climate, aspirate three control flies, one octopamine low fly, and a virgin female a courtship well and record with a video camera for later analysis on the amount of wing extensions, which are the courtship behavior, and lunges, which are the aggressive behavior.

**Results:** Out of nineteen assays with three control, one octopamine low, and one virgin female, the control fly copulated nineteen times. The average number of courtship behavior exhibited by the control flies also far exceeded the octopamine low fly. On average the control flies performed around thirty wing extensions towards the female, while the octopamine low fly on average performed zero wing extensions towards the female. Although when placed in a courtship assay with only octopamine low flies and one virgin female, copulation takes place. Pre-copulation neither type of fly on average performed any lunges, however post-copulation the control fly on average performed around one hundred forty five lunges while the octopamine low did not perform any lunges on average. Post copulation, there is also an increased number of male-male courtship behaviors. In pre-copulation, the number of wing extensions a control fly performed to the octopamine low fly average around two, and the number of average wing extensions an octopamine low perform towards a control average around two. Although post copulation, the number of control to octopamine low rises to an average of around sixteen, the number of octopamine low wing extensions towards control flies averages less than 1, and the controls also begin to court other controls, on average around twelve times.

**Conclusions:** Based on these results, male flies with low levels of octopamine are unable to compete with flies who have normal levels of octopamine in a social courtship setting. Also, flies with normal levels of octopamine begin to exhibit a lot of aggressive behavior post copulation, while flies with normal levels of octopamine do not lunge at all. Post copulation the amount of male-male courtship also increases, and shows control males have a slight preference for the octopamine low males. Overall, octopamine has a significant effect on both courtship and aggressive behaviors in male *Drosophila*.

# **The Utility of *Ustilago bullata* to Control Cheatgrass Invasions**

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## **Introduction**

*Cheatgrass (Bromus tectorum)* is an invasive species of grass found all throughout the intermountain west of the United States, and is controlled by the use of herbicides, tillage, or burning which is damaging to the surrounding environment. Cheatgrass populations exist on MPG Ranch which is conservation property in western Montana. Some populations of cheatgrass are infected with the fungi *Ustilago bullata* (commonly head smut) which prevents the development of seeds in cheatgrass. A better understanding of *U. bullata* and the relationship between the fungi and cheatgrass could lead to the utilization of *U. bullata* as a biological control agent for cheatgrass.

## **Methods**

Some populations of cheatgrass on MPG Ranch are more heavily infected with *U. bullata* than others, which could be explained by differences in the fungal endophyte community of various cheatgrass populations. T-RFLP analysis was used to assess the fungal endophyte community of cheatgrass populations. Significant differences in endophyte communities have been found in cheatgrass samples from MPG Ranch.

## **Results**

The T-RFLP analysis showed differences in the fungal endophyte communities between roots and shoots ( $p=0.002$ ), and site 1 and site 2 showed significantly different communities with  $p$ -values of 0.002 and 0.034 respectively. An analysis of variance (ANOVA) of length of healthy and unhealthy cheatgrass showed significant difference. Those plants infected with *U. bullata* were on average 10 cm shorter than healthy plants. This difference in height between diseased and healthy plants was shown to be significant ( $p<0.001$ ).

## **Conclusions**

Surveys have shown *U. bullata* occurs in many places where cheatgrass is present, except for remote wooded locations. This suggests the pathogen is dispersal limited and inoculations may be necessary in order for cheatgrass infection to reach a level which could be epidemic. Our small-scale survey indicated large variations in *U. bullata* abundance when examined at a meter scale on the ranch. The underlying reasons for this great variability are currently unknown, but our T-RFLP data suggests differences in fungal endophyte communities of healthy and diseased cheatgrass could play a role. Healthy plants harbored, on average, one more endophyte in the roots and shoots than diseased plants. It is possible the presence or absence of specific organisms in cheatgrass populations affects the likelihood of *U. bullata* infection in cheatgrass plants.

## ***4-Isoxazolyl-1,4-dihydropyridines used as inhibitors for multidrug resistance transporters***

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**Introduction:** The development of multidrug resistance in tumor cell has been recognized as one of the major obstacles to successful cancer treatments. Tumor cells in vitro and in vivo can develop multidrug resistance (MDR) to the lethal effects of a variety of cytotoxic drugs, used to treat cancerous tumor cells. The over production of multidrug-resistance transporter (MDR-1) in cancer cells can be thought of as a protective factor for cancer cells, and a cause of MDR seen in cancer cells. Reversal of multidrug resistance is of clinical interest, and MDR reversing agents such a 4-Isoxazolyl-1,4-dihydropyridines have been under intensive investigated.

**Methods:** The new IDHPs were prepared from isoxazolyl-oxazoline using our lateral metalation and electrophilic quenching methodology. The isoxazolyl-oxazoline was deprotonated using n-butyl lithium at low temperature, and electrophilic quenching provided 5-ethyl analog, which was used to make both branched aryl analogs. After the subsequent metalation and electrophilic quenching step, the oxazoline group was deprotonated by methylation to the quaternary salt, reduction with sodium borohydride followed by mild aqueous hydrolysis to produce the corresponding aldehydes. The Hantzsch procedure for branched aryl examples required that the reaction be conducted at moderate pressure in an aerosol dispersion tube. Screening of IDHPs was performed by the Psychoactive Drug Screening Program (PDSP) of NIMH. The PDSP protocol utilizes live Caco-2 cells, which are derived from human colonic epithelium cells which express MDR-1. The assay is based on the passive diffusion of calcein acetoxymethyl ester (Calcein-AM), which is hydrolyzed inside the cell to calcein, which is both fluorescent and negatively charged. MDR-1 can transport non-fluorescent Calcein-AM from cells. The assay measures the increase in calcein fluorescence as a function of time using a FlexStation II fluorimeter in 96 well plates in which cells were preincubated with IDHPs for 30 minutes, upon which time calcein-AM was added to a final concentration of 150 nM. Fluorescence is monitored over 4 minutes, and each assay was performed in quadruplicate, with a cyclosporin control.

**Results & Conclusions:** We now report that 4-Isoxazolyl-1,4-dihydropyridines are effective MDR-1 inhibitors. With the aid of computer modeling a SAR has been developed for further development of second generation 4-Isoxazolyl-1,4-dihydropyridines MDR-1 inhibitors.

# **Analysis of Northern Goshawk Nest Site and Nesting Habitat on the Lewis and Clark National Forest**

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**Introduction:** *With the infestation of the Mountain Pine Beetle through the national forests of Montana, namely the Helena National Forest, Lewis and Clark National Forest, and the Custer National Forest, there is the potential for adverse effects of this infestation on the Northern Goshawk (accipiter gentiles). Through utilization of the National Agriculture Imagery Program (NAIP), and ARC GIS software, as well as nesting habitat and nest site data, identification and analysis of current parameters regarding nest site selection will be determined. This analysis will then be used to identify quantitative parameters that can be used to modify potential habitat that may provide suitable refuge for the Northern Goshawk after Mountain Pine Beetle destroys much of the current nest sites. .*

## **Methods:**

*The protocol set forth in the Northern Goshawk Inventory and Monitoring Technical Guide, August 2006 (B. Woodbridge and C.D. Hargis for Intensive Search and Broadcast Acoustical goshawk surveys was used to perform the surveys of Goshawk nesting habitat and nest sites. The analysis of these parameters was gleaned from a total of twenty-one nest sites on the Lewis and Clark National forest both in the Rocky Mountain Front Range as well as within the Little Belt, Castle, and Little Snowy Mountains. Remote sensing technology (NAIP and Satellite imagery) and GIS software were used to quantify nest area and post-fledging areas.*

## **Results:**

*Data analysis of nest sites show the occurrence of Douglas Fir as the most abundant nest tree (57%), with 90% of the nest trees being alive and branch whorls being the most abundant nest structure (47%). With regards to habitat analysis the comparison of tree species across each territory (measured as tree species present within the post-fledging area) was found to be significant at a p-value of  $8.95 \times 10^{-5}$  while the comparison of tree species within each territory was not found to be significant at a p-value of 0.89.*

## **Conclusions:**

*The data analysis shows similar results regarding nest site and nesting habitat parameters with a number of other studies performed within Montana such as that performed by Hayward and Escano, 1989. Furthermore, based on these parameters a qualitative understanding of goshawk nest site preferences can be deduced and utilized for the recommendation of treatments that can be implemented when goshawk species become further threatened by the Mountain Pine Beetle epidemic.*

## **Macroinvertebrate processing of non-native leaf litter in the Stillwater River**

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**Introduction:** *In stream ecosystems, decomposition of leaf litter from riparian trees is a major component of nutrient cycling. Macroinvertebrates contribute to this by colonizing and processing the litter. When native tree species are replaced by non-natives, litter entering the system changes, and macroinvertebrate use of this non-native litter could be altered. We studied the effects of Russian olive (*Elaeagnus angustifolia*) invasion on nutrient cycling in the Stillwater River riparian ecosystem by comparing rates of macroinvertebrate colonization and processing of Russian olive leaf litter and litter from native narrow leaf cottonwoods (*Populus angustifolia*).*

**Methods:** *We collected leaves from Russian olives and narrow leaf cottonwoods in late September 2011, brought them back to the laboratory, and air dried them for one week. We placed 8 gm of dry leaf material into litter bags, constructing a total of 24 bags for each species. We attached bags to rebar and placed them in the river at six riffle sites. At each site, there were four litter bags for each species. After two, three, four, and five weeks, we collected one litter bag of each species from each site. Within 24 hours of collection, we rinsed sediments and macroinvertebrates from the remaining leaf material. We counted macroinvertebrates and identified them to Family. Using the Shannon-Wiener index, we compared biodiversity of the communities supported by the litter of each species. We also dried the remaining leaf material at 45 degrees Celsius for 72 hours to determine how much leaf material remained and compared the litter mass lost between the two tree species.*

**Results:** *The litter of both species supported a community of macroinvertebrates dominated by stoneflies, mayflies, and caddis flies. Over the course of the study, there was no clear difference in the diversity of the communities supported by Russian olive and cottonwood leaf litter as measured by the Shannon-Wiener index. However, there were more total individuals found in bags containing Russian olive litter, and these same bags also lost more mass than bags containing native cottonwood litter.*

**Conclusions:** *One hypothesis was that Russian olive litter would attract fewer macroinvertebrates and decompose more slowly because of its high concentration of lignin and because its leaves are generally well-defended against herbivores. However, our data supports the alternative that the extra nitrogen content of Russian olive leaf litter attracts more macroinvertebrates and stimulates quicker decomposition.*

# **Investigation of the Room Temperature Solid Matrix Luminescence Spectroscopy of Polycyclic Aromatic Hydrocarbons in Sugar Glasses**

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## **Introduction:**

*Solid matrix luminescence (SML) is an inexpensive technique for the detection of trace levels of organic compounds. The room temperature fluorescence (RTF) and phosphorescence (RTP) of several polycyclic aromatic hydrocarbons were measured utilizing glasses prepared from sugars as a solid matrix.*

## **Methods:**

*Sugar glasses of glucose, xylose, and maltose were investigated. Model compounds investigated included pyrene, 1-hydroxypyrene and trans-stilbene. Trans-stilbene was used as a rigidity probe of the sugar glasses.*

## **Results and Conclusions:**

*RTF of the compound was easy to observe; however, the RTP of both Pyrene and 1-Hydroxypyrene is low. The RTP was significantly improved by incorporating a heavy atom into the glass: sodium iodide (NaI). The optimal concentration of NaI for pyrene phosphorescence was 12%. Pyrene phosphorescence showed a 44-fold increase in the RTP signal at this concentration, and a 10.5-fold increase for 1-hydroxypyrene was observed at 12% NaI. The average phosphorescence lifetime for pyrene with 10% NaI was  $28.8 \pm 4.78$  ms. The effect of the addition of two polymers—polyacrylic acid (PAA) and polyvinylpyrrolidone (PVP)—at a concentration of 1% and 2% was examined for their effect on the rigidity of the glasses. Both polymers produced hard glasses. However, trans-stilbene fluorescence measurements did not reflect an increase in rigidity. Residual water left over from the glass preparation decreases the rigidity of the glass matrix resulting in decrease luminescence intensities. Glucose glasses are the best for measuring the RTF and RTP of the compounds investigated. However, maltose demonstrated an increased RTP for pyrene compared to glucose.*

# **An In Vitro Study of the Use of Curcumin to Inhibit the Proliferation of Malignant Brain Tumor Cells**

*Julia Michels*

*Big Sky High School, Missoula, MT*

## **Introduction:**

*This project was designed in order to explore the use of natural medicines to treat cancers. This project used curcumin, an extract of the cooking spice turmeric, to inhibit the growth of malignant human brain tumor cells.*

## **Methods:**

*All of the following procedures will take place in Dr. Beall's lab at the University of Montana. The lab is a sterilized environment, and therefore requires knowledge of procedures and caution when working with the sensitive materials. The SNB19 cell line is brought up from the frozen stored, then cultured in DMEM until ready to test. They are then removed from the flasks and put into 96-well plates for testing. The curcumin is then made into 1, 10, and 100 $\mu$ M stock solutions in order to create the other solutions for testing. The cells are then treated in a 0- 250 $\mu$ mol/L dosage range for 4 hours, and then analyzed with an MTT assay. The data from the MTT assay was then analyzed with a plate reader.*

## **Results:**

*The MTT assay was replicated 3 times, which produced 3 IC<sub>50</sub>, or half-maximal inhibitory concentration value at which 50% of the cells are viable, and 50% are dead. The three values are as follows:*

*Plate 1: 14.477 $\mu$ M*

*Plate 2: 27.606 $\mu$ M*

*Plate 3: 13.254 $\mu$ M*

## **Conclusions:**

*This study is only a preliminary study, so the only conclusion that can be drawn is that curcumin inhibits the growth of malignant brain tumor cells. Curcumin can cross the blood brain barrier, which makes it a viable option for brain cancer treatments. My hypothesis was supported by the data, in which all three values fell between 10 $\mu$ M and 30 $\mu$ M.*

# **The Investigation of the Biological Properties of the Ayurvedic Plant *Bacopa monnieri***

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<sup>1</sup>Big Sky High School

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## **Introduction**

*Bacopa monnieri* is a wet, creeping herb commonly prescribed in India as an Ayurvedic medicine. It is a relaxing plant, used for treating anxiety attacks and arthritis, relieving certain pains, and treating inflammatory conditions such as cardiac, respiratory, or neurological diseases. Recent Western studies have also showed *B. monnieri*'s ability to treat certain cancers. If *B. monnieri* can inhibit the signal transduction of certain enzymes upregulated during inflammation, cell death, and cancer, then compounds within *B. monnieri* can potentially be isolated and used to treat certain diseases.

## **Methods**

Chloroform and chloroform-alkaloid extractions of *B. monnieri* were separated via normal phase silica gel chromatography. Extractions and their resulting fractions were tested for signal transduction inhibition of caspase-1, caspase-3, and matrix metalloproteinase-3. Fractions which showed the most signal transduction inhibition were continued and broken down through further chromatography separation to work towards isolating a compound. Fractions were also put through proton nuclear magnetic resonance (NMR). When fractions showed the same NMR results, it was assumed a single compound was isolated.

## **Results**

*B. monnieri* fractions had beneficial activity, and showed a continued high percentage of signal transduction inhibition of caspase-1 and matrix metalloproteinase-3. Fractions showed moderate inhibition of caspase-3. By observing the NMR graphs, it was assumed a compound originating within the chloroform extraction was isolated. After running the sample through proton and carbon magnetic resonance, along with a number of other tests, its structure was elucidated. The compound, called betulinic acid, was previously known to exist within *B. monnieri*.

## **Conclusions**

Based on signal transduction tests, *B. monnieri* has the ability to inhibit the signal transduction of an anti-inflammatory enzyme such as caspase-1, an enzyme involved with cell death and neurodegenerative diseases such as caspase-3, and an enzyme involved with cancer and arthritis such as matrix metalloproteinase-3. By using normal phase silica gel chromatography, compounds within *B. monnieri* can be isolated. This study supports betulinic acid is contained within *B. monnieri*, and with further chromatography, other compounds with pharmacological potential can possibly be isolated.

# ***A Special Case of Hedetniemi's Conjecture***

*Demitri Plessas.*

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**Introduction:** In the 18th century the town of Königsberg in Prussia, now Kaliningrad in Russia, was built on both sides of the Pregel River. The river splits and creates two large islands which are connected to the mainland and each other by seven bridges. Mathematicians would share their favorite problem about the town that it seemed no one could solve. Can you walk around the city, crossing each bridge exactly once?

During 1735 in St. Petersburg, Russia, Leonhard Euler answered the question, and doing so created a new branch of mathematics called Graph Theory. He imagined a new mathematical structure of dots (called vertices) and lines connecting the dots (called edges). With his structure, he was able to prove a theorem that showed such a tour of the city is impossible.

Euler's solution to this recreational problem has since been used to optimize shipping paths, snow and garbage removal, bus routing, and mail delivery. The branch of Graph Theory is now a mature mathematical discipline used to represent data structures, optimize combinatorial problems, and define social networks.

In London during 1852, Francis Guthrie was coloring a map of England. While coloring the map, he conjectured that only using four colors, he could color any map so that no two regions that share a border are given the same color, although countries that are kitty-corner can have the same color much like a checker-board.

This conjecture was then put in terms of Graph Theory by putting a vertex in the center of each region and placing an edge between two regions if they share a border. Then the conjecture becomes, "you can color the vertices of a graph of a map with four colors so that no two vertices sharing an edge obtain the same color."

Augustus De Morgan learned about Guthrie's conjecture, and De Morgan helped spread Guthrie's conjecture throughout the mathematical community. After over one hundred years of research into the problem, in 1976 Kenneth Appel and Wolfgang Haken at the University of Illinois at Urbana-Champaign finally produced a proof through the use of 1200 computer hours checking cases.

This problem created the sub-discipline of Graph Theory called vertex coloring, where the common question is, "What is the minimum number of colors needed to color the vertices of a graph so that no two vertices sharing an edge are given the same color?" We call this number the chromatic number.

In 1966, S. Hedetniemi conjecture that for finite graphs, the chromatic number of the product of graphs is equal to the minimum of the chromatic numbers of its factors. This conjecture remains open and the study of this conjecture has created the active subfields of multiplicative graphs and exponential digraphs.

**Methods:** *Through our investigation of the categories of a graphs, a new approach to prove cases of Hedetniemi's conjecture was discovered. This approach uses the internal graph structure to recursively "lift" a required graph morphism.*

**Results and Conclusions:** *We use this new approach to prove a special case of Hedetniemi's conjecture, showing the conjecture holds if one of the factor graphs contains a complete subgraph with size equal to the minimum of the chromatic numbers of the factors.*

# **Characterization of the distribution of miniature inverted repeat transposable elements in the genome of *Oryza sativa***

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*Transposable elements are small sequences of DNA that are capable of reproducing within a host genome. Miniature Inverted-repeat Transposable Elements (or MITEs) are a kind of transposable element. They show attributes of two already documented varieties (type I and type II) but do not neatly fit into either category. A brief look showed that unlike type I or II transposable elements, MITEs appeared to be in or near genes more often than not. That distribution is unlike other transposable elements and would presumably be detrimental to the host organism according to our current understanding of gene expression. To further study MITE location, this project computationally automated the characterization of MITEs in *Oryza sativa*, specifically documenting where individual MITEs were in relation to genes. By analyzing over 2000 unique MITEs, we have shown that 80-90% are in or near genes. Almost all of those MITEs are located in non-coding introns of the genes.*

# ***Wolbachia pipientis* in Montanan Spiders**

Emily Sterbis

Big Sky High School, Missoula, MT

**Introduction** *The purpose of this project is to discover the distribution of the bacterial parasite Wolbachia in Montanan spiders. This will add to the available knowledge on which arthropods are infected with Wolbachia and create a base for further research on the horizontal transmission of Wolbachia from predator to prey.*

**Methods** *93 spiders were collected in Missoula, Montana and identified by family and gender. The DNA from these spiders was then extracted using a kit from Promega. PCR was then performed using two sets of primers, one for the spider DNA (cytochrome oxidase I) and one for the Wolbachia DNA (16S rRNA). The PCR product was then examined using gel electrophoresis.*

**Results** *All of the 93 spiders have been identified by family and gender. Of the 93 spiders collected, 42 have been tested for Wolbachia so far. Of the 42 spiders tested, seven have been infected with Wolbachia. The seven infected spiders were spread throughout four families: Miturgidae with one infected individual, Agelenidae with one infected individual, Araneidae with one infected individual, and Theridiidae with four infected individuals.*

**Conclusions** *Of all the spiders tested, 1/6 of the individuals have been infected with Wolbachia. Approximately 28.57% of the tested families have had infected individuals. While these percentages are lower than hypothesized, only one uninfected family has no more individuals left to be tested. So more infected individuals in more families could be found in the future as DNA testing continues. Future research will continue to test all spiders and search for an explanation of why certain families are infected with Wolbachia.*

# ***Investigating how native species are affected by road obliteration in the Lewis and Clark National Forest***

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***Introduction*** *The U.S. Forest Service is enacting a new travel plan in the Lewis and Clark National Forest over the next several years. Part of this work included an inventory; surveying and mapping of existing roads and trails to be closed and restored to a pre-development state by varying degrees. This project will afforded a valuable opportunity to study how the wildlife is affected by, and responds to, this reconstructive effort.*

***Methods*** *The first phase of this project took two years for preliminary data collecting. Roads and trails identified for closure were selected for study based on different methods of restoration used. Extensive field observations included working with the Forest Service throughout the survey and obliteration operations. Primary data collection techniques used included the following: 1. Plot-quadrant sampling of vegetation growing the first and second years after obliteration. 2. Use of Sherman traps to identify small animal species occupying re-contoured road sites. 3. Monitoring large animal activity before and after road obliteration through the use of motion-sensitive trail cameras. Strictly non-destructive methods were used to gather data while leaving a minimal 'footprint' on the recovering ecosystems.*

***Results & Conclusions*** *Data was compared between the predominant methods of obliteration with or without reseeded, and with or without the use of 'slash' (downed trees as top-cover). Preliminary results indicate that vegetation recovery is slow regardless of whether re-seeding is practiced, but that greater diversity occurs in the absence of re-seeding. Noxious weeds were found regardless of method used. The use of slash is of mixed benefit; rodents took advantage of the increased cover, but occurrences of large animal traffic were significantly reduced. The author timed his own hikes along these roads before and after obliteration and found heavy slash and unstable footing (due to soil scarification) to highly detrimental to travel by large animals. Overall, the lack of a distinct organic layer and the inconsistent compaction and aeration of scarified soil may have far more significant effects on the desired outcome of road obliteration than previously thought. More hypotheses may be worth proposing and testing based on the observations made during this study. This research should be continued over a longer term to provide a better understanding of the impact of road obliteration techniques on native flora and fauna. The information gained could help determine which methods are most effective for future restoration work and avoid less effective or counterproductive ones.*

# **Effects of Allelochemicals on Growth of Bromus Tectorum**

Catherine Witt

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## **Introduction**

*Alien, allelopathic plants become invasive in an environment because the native plants of the region have no past experience or resilience to the allelotoxins in the plant. One example of this process, knapweed, has become invasive in many areas of the United States, costing millions to farmers and home-owners. Allelotoxins not only affect people in the United States, but many U.S. plants have become invasive in other parts of the world, especially in China. The allelochemicals in U.S. plants can be very effective when working on plants of a different region with no past chemical experience. This project strives to determine if allelotoxins in U.S. plants can have an effect on the Eurasian cheatgrass (B.tectorum).*

## **Methods**

*Using Solidago canadensis and Panicum virgatum (both native U.S. plants which have become invasive in Asia) a comparison will be made between lengths of cheatgrass when grown alongside native plants and when grown alone. It will be examined whether either of the selected plants have an adverse allelopathic affect on cheatgrass. Then, compare impact of mixing the native seeds at different ratios, on the germination and growth of cheatgrass.*

## **Results and Conclusion**

*After six weeks of growth, cheatgrass grown with Panicum virgatum was almost half the length of the cheatgrass control. Plants grown with Solidago canadensis were on average longer than the cheatgrass control and exhibited signs of better health. In the ratio analysis, when cheatgrass was grown with an even combination of P. virgatum and S. canadensis, germination was only eight percent. This is considerably lower than the sixty percent or greater germination rates of the other groups.*

*Experimentation supported that cheatgrass grown with Panicum virgatum was greatly inhibited in growth when compared to both the control and the Solidago canadensis group. This is an indicator that further study into the chemical properties of Panicum virgatum could yeild a potential herbicide for cheatgrass. It would also be beneficial to study the impact of combining various plants on the growth and germination of cheatgrass. Even slowing the germination could provide an avenue for native and crop plants to gain the upper hand in growth.*

## **Posters**

### **Conserved protein sequences in *Drosophila* TGF- $\beta$ ligands and receptors**

*Luke C. Bates*<sup>1</sup> and *Phillip A. Jensen*<sup>2</sup>

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*A ligand is a molecule that binds to a specific receptor, resulting in a signaling cascade of events at the sub-cellular level. In fruit flies (*Drosophila*) conserved protein sequences have been identified in both a ligand and its receptor. The twelve species of *Drosophila* have been categorized into three groups based on the lengths of the conserved regions of amino acids in the receptors. Intriguingly, each group has a unique, conserved sequence of amino acids in the corresponding ligand. A current hypothesis states that the length of the conserved region in the receptor will determine which of the three slightly different forms of the ligand will bind effectively. The primary goal is to genetically alter the conserved region of the receptor in *Drosophila melanogaster* to mimic the lengths of the conserved regions in the other two groups and measure binding efficiency to ligands from each group. These experiments aim to determine if the lengths of the conserved regions in the receptors are crucial for ligand binding.*

## **Scanning the Horse Genome for Miniature Inverted Repeat Transposable Elements**

*Erin N Burns and Mark T Osterlund. Department of Biology, Rocky Mountain College, Billings, MT*

*A MITE is a sequence of DNA that moves, appears many times in the genome, and is made of terminal inverted repeats (TIRs) flanking sequences of unknown importance. Directed repeats, which occur directly upstream and downstream of the TIRs, are evidence of the MITE's past movement. These characteristics are universal of all MITEs identified in eukaryotic organisms. As a student in both the biology and equestrian programs at Rocky Mountain College, I am working to identify MITEs in horses through the recognition of their described characteristics. I have used automated computational algorithms, developed by the Rocky Mountain College computer science program, to scan the Equus caballus genome for possible MITEs. I am in the process of manually analyzing the 60,000 putative MITEs for the necessary characteristics. Once a MITE is verified we will document its locations and distributions throughout the genome. We can then use that information in conjunction with donor horse pedigrees and DNA samples to identify the genetic relationship between different horses and horse breeds.*

## **Demonstrating Ligand-Receptor Specificity in the PDGF/VEGF Signaling Network in *Drosophila***

Britney Cheff<sup>1</sup>, Kayla Baich<sup>1</sup>, Wyatt Wilson<sup>2</sup>, Philip A. Jensen<sup>1</sup>, and Andy Wildenberg<sup>2</sup>

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*Cells in multicellular organisms must communicate to ensure proper growth and development by sending signals that bind receptors that relay a message. Different signals bind different receptors to communicate different messages to cells. For example, the Activin pathway in *Drosophila* contains four ligands and three isoforms of a single receptor, and different ligand-receptor signaling pairs provide specificity in different tissue types. Because this signaling mechanism employs three different receptors from a single gene it is an efficient way for the cell to sort related signals. To determine if this efficient mechanism is used by other cell-cell signaling pathways in *Drosophila*, an algorithm was developed to search the genome for single receptors that receive multiple related signals. The algorithm identified PVR (PDGF/VEGF receptor), which also has multiple isoforms. Of the isoforms, three are well annotated; these three isoforms all differ in their ligand-binding domains and in the locations of Immunoglobulin (Ig) domains. Intriguingly PVR also has three known ligands: Pvf1, Pvf2, and Pvf3. Here we propose a plan to perform wing assay experiments that are similar to experiments done with the Activins in order to classify specific ligand-isoform interactions. Such experiments could ultimately determine if this signaling-sorting mechanism is common in cell-cell signaling pathways in *Drosophila*.*

# **Mutating Conserved Amino-Acid Residues Crucial for Ligand-Receptor Binding and Specificity in the Activin Signaling Pathway in Species of *Drosophila***

Samantha J. Dietz and Philip A. Jensen, Department of Biology, Rocky Mountain College, 1511 Poly Dr., Billings, MT 59102

*Cells communicate using signaling pathways. Each signal, or ligand, interacts with a specific receptor in a manner similar to a lock-and-key mechanism. Ligands and receptors evolve, or coevolve, over time. 12 species of *Drosophila* have been grouped into 3 sub-groups based on characteristics of their ligands and receptors in the Activin signaling pathway. Ligands within each group of species have a unique pair of similar amino acids within close proximity to one another. The first group of 5 species have H and R in common, the next 3 species have Q and K in common, and the last 4 species have K and A in common. Also, the receptors within each group have a unique number of amino acids in a loop that determines the receptor's shape. The receptors in the same three groups also follow a pattern: the first 5 species have a 12 amino-acid loop, the second group of 3 species have a 13 amino-acid loop, and the last group of 4 species have a 14 amino-acid loop. We hypothesize that these regions of the ligands and receptors are crucial for specific ligand-receptor binding.*

*This hypothesis can be tested by mutating the amino acids in the ligands from group 1 to look like those from group 2 or 3. For example, I will mutate the H-R sequence in a ligand from group 1 to the Q-K sequence found in group 2, and see if this mutated ligand will still interact with the original receptor found in group 1. Reciprocal experiments will also be conducted to see if the newly mutated ligands will interact with receptors from other groups. These experiments will help determine how ligands and receptors have mutated over time and might therefore determine how a lock-and-key mechanism can evolve.*

## **Regionalizing MITEs**

Jacob Downs<sup>1</sup>, Mark Osterlund<sup>2</sup>, Andrew Wildenberg<sup>1</sup>, Andrew Scott<sup>2</sup>

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**Introduction:** Transposable elements (transposons) are units of DNA that move from place to place within a genome. Transposons are categorized as either Class I or Class II elements depending on how they move. MITEs (Miniature Inverted-Repeat Transposable Elements) are distinct types of elements that do not clearly fit into one class or the other. The means by which MITEs move is not well understood, though one clue may be in the documented association between MITEs and genes.

**Methods:** We have developed a program that explores this association by identifying MITEs that occur within genes in the rice genome. The program locates known MITEs with the BLAST search algorithm, retrieves the flanking sequences around each MITE, and then searches for these sequences in an EST database. This process allows us to identify if a MITE is near an exon, which indicates that it is likely in or near a gene.

**Results & Conclusions:** Preliminary results suggest that MITEs occur predominantly in genes, but more tests are needed to verify the accuracy of these results.

## ***The characterization of BH3I-1, an inducer of mammalian apoptosis and an inhibitor morphogenesis in Candida species***

*Mandi Graham, Joy Goffena, David K. Butler, and Kurt A. Toenjes  
Montana State University Billings*

*The switch between yeast and filamentous growth forms and the corresponding changes in protein complement are important for the virulence of Candida albicans, an opportunistic fungal pathogen of humans. Previously, in a large-scale screen of small organic molecules, we found that several molecules with different modes-of-action in non-fungal cells specifically inhibited starvation-induced hyphal growth in C. albicans without affecting cell viability or budded growth. We have screened this group of molecules for the ability to inhibit the switch to filamentous growth forms in conditional *tup1Δ*, *nrg1Δ*, and *rfg1Δ* mutants. The *TUP1*, *NRG1*, and *RFG1* genes normally act to repress hyphal growth. We have found that BH3I-1, an inducer of mammalian apoptosis, inhibits the filamentous growth transitions. Conditional *tup1Δ* MET: *TUP1*, *nrg1Δ* MET: *NRG1*, and *rfg1Δ* MET: *RFG1* strains were created that contained GFP (Green Fluorescent Protein) under the control of the hyphal specific promoter of *HWP1*. When these strains were grown under conditions that repress the MET promoter and in the presence of 100μM and 150μM BH3I-1, BH3I-1 repressed the induction of *HWP1*-GFP in all the mutants. This suggests that BH3I-1 inhibits the induction of *HWP1* expression downstream of these negative regulators and yet upstream of *HWP1*.*

# ***Circadian Influence on Learning and Memory***

*Josi Herron<sup>1</sup>, Bayara Chuluun<sup>2</sup>, Damien Colas<sup>2</sup>, Olivia Jew<sup>2</sup>, and H. Craig Heller<sup>2</sup>*

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## ***Introduction***

*It is known that performance in several learning and memory tasks is under the circadian influence. Through its simple design and flexibility, the novel object recognition (NOR) task is used frequently to quickly assess short-term and long-term memory in mice. The task takes advantage of the innate tendency of rodents to explore novel objects more than familiar ones. The purpose of the current study was to assess how NOR performance varied across the 24-hour circadian cycle.*

## ***Methods***

*By manipulating the training/recognition delay period, we sought to understand the circadian influence in NOR performance. Optimal points of NOR performance were characterized to provide a foundation for future studies that require the use of NOR as a learning task. Learning indices (time spent with novel object/ total exploration time x 100%) were measured and optimal points were defined as those with statistically significant differences ( $p < 0.05$ ) in learning between training and recognition.*

## ***Results and Conclusions***

*Results indicated that optimal points of NOR performance were when training was conducted at the beginning of the light period and recognition was conducted 12 hours later, and also when training was conducted at the beginning of the dark period and recognition was conducted 24 hours later. Further investigation of these optimal points of NOR performance needs to be conducted in order to fully understand the circadian influence on recognition memory in mice. We hope that this research will help in the understanding of the circadian influence on recognition memory.*

## ***Identification and function of the Leptospira biflexa batD protein revealed by means of FIAsh dyes***

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**Introduction:** *Leptospira*, a pathogenic spirochete known for its slenderness and unique flexibility, is the causative agent of the bacterial zoonotic disease Leptospirosis. The mechanism by which leptospire cause disease remains unknown. Analysis of the behaviors and functions of *Leptospira* proteins is highly important to help understand and facilitate control of Leptospirosis. Such analysis can be done by means of Fluorescein Arsenical Hairpin (FIAsh) dyes, fluorescent molecules that can be bound by a small amino acid sequence added to a protein of interest. The genetic fusion of this dye to a protein allows for labeling and detection of the protein in living cells, making it possible to determine the structure and function of the protein. The FIAsh dyes method is especially suitable for labeling target proteins in particularly small bacterium such as *Leptospira*.

**Methods:** To demonstrate the use of this method, the *Leptospira biflexa* Bacteriodes Aerotolerance (*batD*) protein was used as the target protein.

**Results & Conclusions:** Successful use of the FIAsh dyes approach in live spirochetes reveals a promising way to examine other *Leptospira* proteins to help develop a better, more complete understanding of *Leptospira* and how it causes disease, in addition to examination of other small bacterium.

## ***Rebuilding the Gulliver Transposase from Chlamydomonas reinhardtii***

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*Class II transposable elements are sequences of DNA that have terminal inverted repeat (TIR) sequences and a transposase gene between the TIRs. The transposase protein recognizes the TIRs and cuts out the transposable element to move it elsewhere in the genome. There are similar, small transposable elements called MITEs (miniature inverted-repeat transposable elements) that have TIRs but do not contain a gene for transposase. I am interested in a specific Class II transposon in Chlamydomonas reinhardtii, called Gulliver, and a similar family of high-copy MITEs we call Gulliver-Like. Gulliver and Gulliver-Like MITEs have identical TIR sequences. I have identified 112 highly conserved copies of the MITE and I am currently trying to express the transposase gene from a full-length Gulliver transposon to determine if it can interact with these MITEs' TIRs. The ultimate goal is to determine if the Gulliver transposase is responsible for the movement of Gulliver-Like MITEs.*

# **MITE Detection Using Hidden Markov Models**

David Lindenbaum  
Billings West High School

**Introduction:** MITEs (Miniature Inverted-repeat Transposable Elements) are small transposable elements that differ from both Type 1 and Type 2 transposable elements. They are characterized by a high copy number and the presence of terminal inverted repeats (TIR) and directed repeats. Though MITEs are estimated to make up a significant part of many genomes, relatively few MITEs have been identified, there is a need for a reliable general method for identifying them, and it is not even known if all species have them.

**Methods:** A Hidden Markov Model (HMM) was developed to efficiently identify MITEs by finding TIRs separated by an appropriate distance. In order to model inverted repeats on opposite ends of a sequence, the HMM was modified to use column vectors of a matrix  $M$  as its observation sequence, where  $M(i, j) = 1$  if the test sequence at  $seq(i)$  is the complement of  $seq(j - i)$  and 0 otherwise, for  $i=[0, len]$  and  $j=[len/2, 3*len/2]$ . Effectively,  $M$  encodes, for each possible fold of the sequence onto itself, which nucleotides will align with their complement.

**Results:** When queried with a set of about 58,000 sequences from *Oryza sativa* identified as having a high copy number in the genome, the model marked about 3,360 of them as likely being MITEs. Preliminary analysis shows that a large number of these results are likely newly identified MITEs. The identified MITEs have an average sequence length of 200 and an average TIR length of around 13. The TIRs show an average of 84.6 percent preservation. These statistics are approximately equivalent to known MITEs.

**Conclusion:** Because the algorithm doesn't use any information or assumptions that are particular to *Oryza sativa*, this process should be able to discover MITEs in any species.

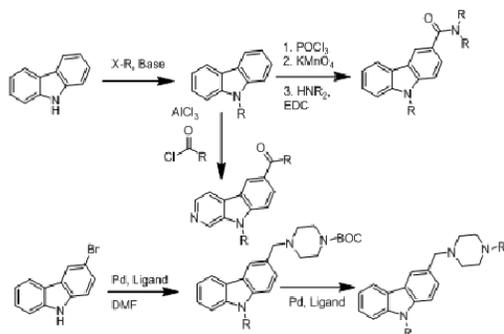
# Design and Synthesis of Dual Modulators of Cannabinoid Receptors and Serotonin Transporter

Steven McDaniel, Ravil Petrov, Fernando Cardozo-Pelaez and Philippe Diaz

Core Laboratory for Neuromolecular Production, Dept. of Biomedical and Pharmaceutical Sciences, The University of Montana, Missoula, Montana 59803

**Introduction:** The development of novel dual modulators of cannabinoid receptors and serotonin transporter that could provide insights of the pharmacology of these targets. These could develop into medical treatments.

**Methods:** The carbazole scaffold is commercially available that is easily modified by alkylation of the nitrogen. Then the carbazole can either undergo functionalization by either Friedel-Craft acylation or formylation, followed by oxidation and amidification (Scheme 1). The bromo carbazole scaffold is also commercially available which can be modified by a suzuki like coupling with trifluoroborates. Then can be further functionalized by deprotection of the boc then a hartwig-buchwald reaction.



**Results:** A novel series has been synthesized and preliminary results show that nanomolar binding can be achieved for cannabinoid receptors and serotonin transporter

Compound	CB1	CB2	SERT
	166 nM	48 nM	321 nM
<b>2</b>	388.7 nM	1405 nM	2983 nM

**Conclusion:** It is possible to design and synthesize dual SERT and CB<sub>1/2</sub> modulators

# **Using feather genetics to determine the relatedness between goshawks in the Lewis and Clark National Forest**

Ashly Pezel, Nate Bickford, and Diane Lund

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**Introduction:** *The ultimate goal of this research is to determine the degree of isolation between goshawks in the Lewis and Clark National Forest. Instead of using traditional mark and recapture methods, a genetic mark and recapture method has been chosen. Molting of feathers begins in the spring and most of these molts are dropped in their nest areas, even when nesting fails, making this a probable means of non-invasively sampling the populations.*

**Methods:** *Molted feathers have been collected over the past several years from nesting sites throughout the Lewis and Clark National Forest. So far, this has turned into a study to solidify methods for isolation of DNA from the collected feathers, which has proven to be difficult depending on the type of feather used, the condition in which the feathers are in, and how they have been stored.*

**Results & Conclusions:** *Many practice rounds, as well as some on the collected feathers, have been performed, which involves cutting the calamus tips of the feathers into small pieces and using the Qiagen DNeasy® Tissue Kit to lyse the cells, digest the proteins, and isolate and precipitate the DNA into solution. After DNA extraction, this DNA will be put through rounds of polymerase chain reaction to amplify certain locus specific regions of genes in order to determine the relatedness between these sampled goshawks in the Lewis and Clark National Forest.*

# **Characterization of *tnrc4*, XI.25952, and XI.8933 in Early Neural Development**

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<sup>3</sup>Department of Cell Biology and Neuroscience, Montana State University, Bozeman, MT

## **Introduction:**

*Early embryonic development of the nervous system is relatively similar in most vertebrates. In most amphibians, including our model species *Xenopus laevis*, primary neurulation is responsible for formation of the neural tube. The genes *tnrc4*, XI.8933, and XI.25952 are all up-regulated by the transcription factor *Zic1*. Studying their expression patterns will lead to a better understanding of their roles and the role of *Zic1* in early neural development.*

*My research is to: 1) determine the expression patterns of *tnrc4*, XI.8933, and XI.25952 in different stages of development relevant to neurulation (i.e. stages 15-21); 2) explore the effects of over expressing and inhibiting expression of these genes on neural development; and 3) conclude the roles of these genes during embryonic development and relate this information to other species, such as humans and mice.*

## **Methods:**

*The specific locations of gene expression were determined using in situ hybridization, a process that stains the area in which a gene is expressed during a particular stage of development, in order to determine variances in gene expression throughout embryonic development. The stained embryos were then evaluated in whole mount and cryosections. At this point, a detailed map of expression patterns has been determined for the three genes in question during relevant stages of neural development for both whole mount and sectioned embryos. Currently work is focused on the up and down regulation of the genes through the use of morpholino oligonucleotides to help determine the roles of these three genes in the process of neurulation. The process of inhibiting gene translation will be achieved by injecting *Xenopus laevis* embryos with morpholino oligonucleotides specifically designed to the *tnrc4* and XI.25952 genes.*

## **Results:**

*My results suggest that *tnrc4* and XI.25952 may be expressed in regions that are closer together in the neural plate, while XI. 8933 may be expressed further away from the midline of the neural plate. These initial results suggest that *tnrc4* and XI.25952 may play a role in development of the neural plate, while XI.8933 might be a part of neural crest formation.*

## **Conclusions:**

*Understanding the process of neurulation is crucial in developing prevention and treatments of disorders that occur during neurulation that impact individuals, such as spina bifida and anencephaly. The future of this work will focus on determining the roles of these three genes during development.*

# Flax Seed Biodiesel

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*Flax is a plant mainly grown in North Dakota and Minnesota. Flax oil is popular in the health food industry to lower cholesterol, treat arthritis, and use as a laxative. This research explored flax seed oil as potential biodiesel fuel. Flax seed oil (linseed oil) was converted through the transesterification process. After the conversion to biodiesel, the fuel was washed and sent to MSU-Havre for analysis. The flax seed biodiesel was analyzed for flash point, cloud point, cold filter plugging point, copper strip corrosion, sulfur content, carbon residue, and water and sediment. The flax biodiesel passed the biodiesel flashpoint standard, EPA sulfur standard, and carbon strip corrosion standard. It had a cloud point of  $-2^{\circ}\text{C}$  and cold filter plugging point of  $-12^{\circ}\text{C}$ . It did not pass the carbon residue or water and sediment standards. However, these problems can be corrected. Based on this research, flax seed oil is suitable for biodiesel fuel.*

## **Rainwater in Central West Montana**

Mike Shirley

**Introduction:** During the summer of 2011, the Montana Department of Agriculture conducted a study of rainwater in central west Montana to investigate the effects of pesticide on rainwater. Three locations were selected; Macdonald Pass (16 miles west of Helena in the Helena National Forest), a location just outside of Townsend (34 miles southeast of Helena), and a location just outside of Toston (45 miles southeast of Helena). Samples were sent to the Montana Department of Agriculture Analytical Lab and analyzed for pesticides.

**Methods:** Precipitation collectors were deployed in the three locations above. They consisted of a metal funnel fixed with sheet metal to increase surface area. The funnels drained into amber glass bottles, and rainfall was tracked using websites linked to nearby weather stations. Samples were collected no later than 48 hours after a rain event, and sent to the analytical lab in Bozeman.

**Results and Conclusion:** All samples obtained contained pesticides in varying amounts. So far a trend in pesticide concentration has not been observed, however statistical analysis is being conducted to determine if proximity to agricultural activity increases the mean concentration of pesticides.

# ***Using Otolith Microchemistry to Determine the Life History of Walleye, Sander viterus, found in the Missouri River and Holter Lake***

Andrea Spake

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**Introduction:** *Walleye, Sander vitreus, is a commonly found fish in central Montana. The life cycle of these fish is thought to be highly predictable. Recently claims have been made that Walleye found in Holter Lake are making it past the dams and living in sections of the Missouri River that are thought to be premiere trout habitat. To determine if these claims are at all feasible determining the life cycle and history of fish found below the dams.*

*Otolith microchemistry has become a widely used method for determining the age and life histories of marine fishes. We propose that these techniques can be utilized in a freshwater habitat as well. By taking samples of larval fish soon after spawning and continued to the juvenile stages, otolith microchemistry can be looked at to look for unique traces in the early stages of life.*

**Methods:** *To sample for larval fish a time frame was selected based off of previous years spawning's. The months of May and June have extensive sampling in which two time periods will be looked at. A morning sampling, one that is done before the sun rises, and a night sampling, one that is concluded before the sun sets will be tested weekly. Four different sites have been chosen along the Holter Lake reservoirs and the Missouri River. For these sites sampling will take place along both banks for a time period that will be determined during the first week of sampling. It is thought that 10-12 minutes will be a sufficient amount of time. Samples will be dyed to determine living matter placed in ethanol and taken to the lab to be sorted through. Samples that are Walleye will be removed and dissected to remove the otoliths. The otoliths will then be mounted on a glass slide in epoxy and sent to an external lab to be tested.*

**Results:** *At this time no results are available due to sampling beginning in May of 2012.*

**Conclusion:** *No information can be concluded at this time.*

# **Characterizing Antibiotic Resistance of *E. coli* Isolated from the Little Big Horn River**

Sydni Racki<sup>1</sup>, Greg Spina<sup>1</sup>, Lance Yellowmule<sup>1</sup>, Andrew Linqvist<sup>1</sup>, Steve Hamner<sup>2</sup>, Cristi Hunnes<sup>1</sup>

<sup>1</sup>Rocky Mountain College, Billings, MT

<sup>2</sup>Montana State University, Bozeman, MT

*The purpose of this research was to test antibiotic resistance in E. coli. The E. coli samples were collected from the Little Big Horn River, near a Concentrated Animal Feed Operation (CAFO) drainage ditch and from the Black Bridge area. The Little Big Horn River is located on the Crow Indian Reservation in Southeast Montana, and the collection areas were about 50 miles north of Crow Agency. Antibiotics are used in the feed that is given to cattle to keep the animals healthy and to promote growth; however, the bacteria that are naturally in the cattle's digestive system are being selected for resistance to the antibiotics that are used in the cattle. After natural processes occur, the resistant bacteria are deposited into the environment and with help from other animals and the weather, the bacteria enter the river and spread to down-stream areas. The E. coli were analyzed for antibiotic resistance using Kirby-Bauer disc-diffusion assays. The antibiotics that were used were ampicillin, cefazolin, streptomycin, nalidixic acid, piperacillin, gentamicin, chloramphenicol, amikacin, ciprofloxacin, tetracycline, ceftazidime, and sulfamethoxazole with trimethoprim.*

*The majority of the E. coli samples were susceptible to the tested antibiotics. One E. coli sample out of nineteen from a Black Bridge sampling area showed intermediate susceptibility to ampicillin. Four E. coli samples out of six from a CAFO sampling area showed resistance to streptomycin and tetracycline, and three E. coli samples out of ten, from a different CAFO sampling area, showed resistance to streptomycin and tetracycline. In addition, one E. coli sample out of twelve collected from the CAFO drainage ditch, showed resistance to ampicillin, and cefazolin.*

# **Selective Predation on Ichthyophonus-Infected Pacific Herring**

Tim Taylor <sup>1</sup>, Paul Hershberger <sup>2</sup>, Lucas Hart <sup>2</sup>, Jake Gregg <sup>2</sup>, Arild Folkvord <sup>2</sup>, and Knut Vollset

(1) University of Great Falls Biology Department, Great Falls, MT 59405, (2) U.S. Geological Survey – Marrowstone Marine Field Station, Nordland, WA 98358 (3) Department of Biology, University of Bergen, N-5020 Bergen, Norway

**Introduction:** *Ichthyophonus hoferi* is a parasite that is prevalent in many fish species in the Pacific Ocean. The purpose of this experiment was to identify if predacious fish species are targeting diseased fish which further spreads the parasite. A previous experiment directly related to the present one was conducted by Knut Vollset. In his experiment, the prevalence of infected prey was 74%, deeming the hesitant results. This experiment solidifies the infection status to 100% to achieve sound results.

**Methods:** Selective predation of diseased Pacific herring (*Clupea pallasii*) was studied in an indoor mesocosm. Lingcod (*Ophiodon elongates*) were allowed to prey on schools of herring that were approximately a year in age; half being infected with *Ichthyophonus hoferi*, the other half injected with a Phosphate Buffer Solution. Once 30-50% of the population was consumed, the leftover herring were observed to determine how many *I. hoferi* injected herring were left. Selection index was calculated from the composition of infected versus non-infected prey. In order to further demonstrate selective predation on infected prey, a large scale experiment was also ran over a week-long period with 100 infected herring comingled with 100 PBS-injected herring with increased number of predators. T-tests were ran to statically observe selectivity—a Chi Squared test was also ran to test whether or not the trials were influenced by selectivity. **Results:** The first experiment calculated a Selection Index of 0.428. A total of 45 infected herring and 41 PBS-injected herring were consumed throughout the trials. The Chi squared test signified that each trial was no influence on selectivity through how the trials were conducted. The second experiment 30 infected herring and 27 PBS-injected herring were consumed over the week-long period. Applying a T-test to the results obtained a p-value of 0.754.

**Conclusions:** These lab based predation trials were unable to demonstrate selection of predators for prey that were diseased with *Ichthyophonus hoferi*. Upon sampling the Lingcod used in the trials, all of the Lingcod who consumed an infected herring also became diseased.

## The Search for MITEs

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**Introduction:** Miniature Inverted-repeat Transposable Elements (MITEs) are a subset of transposons that are often replicated thousands of times in a chromosome, but their origin, function, and means of replication are poorly understood. A preliminary step to understanding MITEs is their identification and cataloguing.

**Methods:** My work has focused on the computational identification of potential MITEs by exploiting their high copy rate. I do this by creating an index of small groups of nucleic acids and identifying overrepresentation. These potential MITEs are then passed through a filter for other structural properties.

**Results:** Results are promising though low entropy regions of genomes often generate false positives. This program is currently being used to identify MITEs in *Chlamydomonas reinhardtii* (green algae) and *Equus caballus* (horse).

**Conclusions:** The method is promising though further work could be done to restrict false positives.

# **G-4 Quadruplex DNA as an Anti-Cancer Target**

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## **Introduction:**

*The focus of this project is to synthesize specialized molecules that bind to G-quadruplex (G4) DNA through multiple interactions including ionic bonding with cations. Stabilization of G4 will facilitate repression of cellular replication in cancerous tumors. G4 is a DNA structure with unique folding and is found in the human telomere and promoter regions of many oncogenes.*

## **Methods:**

*Cations such as K<sup>+</sup> and Na<sup>+</sup> are often trapped in the cavity of G4 when it takes on its folded motif from the normal double stranded conformation. This cavity along with the cations may provide a binding site allowing for the stabilization of the G4 and in turn preventing replication of cancer cells. Anthracenyl isoxazolyl amides (AIMs) represent a new class of antitumor agents developed in our laboratories and show potential to bind G4. Stabilization of G4 limits its structural flexibility and inhibits replication of oncogenes. AIMs are a good candidate for stabilization of G4 because they contain an isoxazole moiety which anchors the anthracene in a fixed position, allowing  $\pi$ - $\pi$  stacking between the G4 cavity and anthracene. Our current lead compound, to which current work is devoted, has shown promising anti-cancer activity, data to be published. Along with increasing activity with structural variations, focus also pertains to streamlining the synthesis and increasing yields.*

## **Results:**

*The anthracenyl isoxazolyl amides have been a focus of our research group for many years. They have provided us with multiple lead compounds and now compounds which have been shown, by the National Cancer Institute, to be more effective against cancer than some treatments currently in use today, data to be published.*

## **Conclusions:**

*Based on current data, to be published in collaboration with Howard Beall, we propose that attaching an aryl group, via an electronegative linker, to current lead molecules will increase the molecules' efficacy and aid in more favorable pharmacokinetic parameters. We will attach N or O, with an aryl group, to the C-10 of an N-(5-(bis(3-(dimethylamino)propyl)carbamoyl)-1-methyl-1H-pyrrol-3-yl)-3-(10-chloroanthracen-9-yl)-5-methylisoxazole-4-carboxamide and predict that the lone pair of electrons will increase the affinity of the novel AIM to bind the cation in the cavity of G4 through dipole and van der Waals interactions. To synthesize this molecule, Buchwald-Hartwig palladium-catalyzed cross-coupling reaction will be used. This approach will allow us to work from a common AIM precursor to which we can add various substituents. We will then identify the AIM variants that bind G4 and test their ability to repress replication.*

## Research into Cysteine Shaping of Trans-membrane Receptor Proteins

Wyatt Wilson. Senior, and Brittney C. Junior, and Andy W Faculty and Phil J. Faculty.

*Advanced research into extra cellular domains of trans-membrane receptors could yield advancements in the understanding of receptor shaping. In particular focusing on the Cysteine markers of the extra cellular region of the receptor could introduce an insight into a wide area of bio-informatics and biology research not just limited to the common fruit fly. In order to assess these potentials accessing the online storage database FlyBase and combining it with TMHMM, and advanced prediction software aimed at trans membrane helices in proteins. From there a solution similar to the Levenshtien Distance Solution is applied to determine high entropy regions and candidate protein sequences for study. For most of the resolved data that passes screening for known shape differentials, short contained limited entropy regions provide a promising outlook into Cysteine shaping of receptors. However, the question of the effect of tail or head insertions of chains of different proteins from the same gene yields another potential questions. However interpretation of the data, and different restructurings of the Cysteine markers due to varied and differing orders of insertion, deletion or substitution operations between sequences adds uncertainty to the mix that will take further experimentation to eliminate. Proving that Cysteine binding of the extra cellular domains shapes receptors opens and entire field for large scale of statistical study. Even if Cysteine are proven to be statistically irrelevant their elimination eliminates a highly considered possibility from further contention in future studies.*